STABILIZATION OF ARID SANDY SOILS USING CYANOBACTERIAL INOCULANTS

FINAL REPORT

STABILIZATION OF ARID SANDY SOILS USING CYANOBACTERIAL INOCULANTS

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by

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ABSTRACT

A total of 140 meter square plots were established at Yuma Proving Ground in a sandy area west of Hwy 95. Of these, 100 were set up in 1994, and consisted of 50 foot-trampled plots and 50 undisturbed plots. The additional 40 plots were set up in 1995, and received vehicular disturbance. Half of the plots established in 1994 were inoculated with pelletized *Microcoleus* in February of 1995. Half of the vehicular-disturbed plots were inoculated with pelletized *Microcoleus* in February of 1996.

In May of 1995, the original 100 plots were sampled for algal abundance, which was estimated using chlorophyll *a* concentrations and the most probable number method. Both methods demonstrated significant differences within the disturbance treatment, with trampled plots having lower algal abundance than undisturbed plots. Both methods were not able to pick up significant differences between inoculation treatments when all data were used in a full multifactor ANOVA model, although means were consistently higher in inoculated plots than in uninoculated plots. When just trampled plots were analyzed with a nonparametric alternative (Mann-Whitney test), the inoculated plots had significantly higher chlorophyll *a* concentrations than in uninoculated plots.

These results are very promising, and we are optimistic that application of pelletized cyanobacteria may be able to stimulate recovery of the algal component of microbiotic crusts. Our enthusiasm is somewhat guarded at Yuma Proving Ground, however, because the densities of algae in undisturbed soils at this site are lower than densities reported in other arid and semi-arid sites.

PERSONNEL AND RESEARCH OBJECTIVES

This report covers research on use of pelletized cyanobacterial soil amendments at a field testing site at Yuma Proving Grounds, Yuma, Arizona. The work began 9 May 1994. The current contract ended 31 December 1995, although continued monitoring of the site past this date is being conducted with funds from another source (USACERL). The principal investigator on this project was Jeffrey R. Johansen, with chief collaborators being Larry St. Clair (BYU), and Valerie Flechtner (JCU). Bruce Webb (BYU) characterized the soils. Valerie Morrill was our point of contact at Yuma Proving Grounds. Stuart Sanderson assisted with vascular plant characterization. Undergraduate student helpers at John Carroll University included: Nicole Trombetta, Susan Okuley, Marissa DeNoble, Mike Payne, and Julie Sray.

The initial research objectives of this study were:

A. Establishment and characterization of study site at Yuma Proving Grounds, Yuma, Arizona (May 1994).
   1. Visit Yuma Proving Grounds, Arizona and chose one hot desert study site on the base.
   2. Characterize the vascular plant community using the nested frequency method.
   3. Characterize the lichen community using the nested frequency method.

4. Characterize the algal community using standard dilution plate methods to quantify cyanobacteria and chlorophytes, and identify algae to determine similarity of crusts to the Dugway site.
5. Characterize soils (texture, pH, chemistry).
6. Trample plots assigned randomly to this treatment. Since the sites are distant from the researchers and are in more sandy, fragile soils, perform a single trampling treatment .
7. Sample for algae. Algal biomass to be estimated by measuring chlorophyll *a* and direct quantification of cyanobacterial biovolumes using epifluorescence microscopy.

B. Produce cyanobacterial inoculum (June-November 1994)

C. Apply cyanobacterial inoculum to control and treated plots (October-November 1994).

D. Resample algae (May-June 1995)
1. Estimate algal biomass by measuring chlorophyll *a*.
2. Estimate cyanobacterial biomass by direct quantification of cyanobacterial biovolumes using epifluorescence microscopy.
3. Test sedimentation and runoff using an artificial rain device.

ESTABLISHMENT AND CHARACTERIZATION OF STUDY SITE

Yuma Proving Grounds was visited for the first time on 12-17 May 1994. Seven sites in the area were sampled for soil algae (Fig. 1). Latitude, longitude, and a short description of these sites are given in Table 1. A study site was eventually chosen in a very sandy area slightly east of Hwy 95. The site was called the macroplot (MACRO, Fig. 1, Table 1), and contained 100 permanent plots, consisting of 25 blocks of 4 plots (Fig. 2). In each block, the following treatments were randomly assigned to the four plots: 1) trampled, to be inoculated, 2) trampled, not to be inoculated, 3) undisturbed, to be inoculated, and 4) undisturbed, not to be inoculated. Trampling was accomplished by vigorous foot traffic, including stomping and kicking activity. Ten of the blocks were randomly designated as enumeration plots, i.e. plots from which soils would be taken for chlorophyll *a* and epifluorescence analyses. The other 15 blocks were designated as sedimentation plots, for study of the erodibility of the soils.

The vascular plant community of the macroplot site was characterized using the nested frequency quadrat method. A metal-frame 0.25 m^2 quadrat with 0.125, 0.0625, and 0.0125 m^2 subquadrats to obtain frequency of vascular plant species was used. The frame also had four points which were read to obtain a direct estimate of cover for both vascular plant species and general cover classes of litter, rock, and sand. Each of the 100 plots had a single nested quadrat scored in the middle of the plot. Frequency of occurrence for vascular plant species was calculated based on all 400 quadrats. The plants on the site were fairly typical of sandy-soiled Sonoran desert communities (Table 2). The dominant taxa based on frequency data were Mediterranean Grass (*Schismus barbatus*), Cryptanth (*Cryptantha* species), and Stevia Dusty-maiden (*Chaenactis stevioides*), while those dominant in terms of cover were Stevia Dusty-maiden and Creosote (*Larrea tridentata*). The differences between frequency data and cover data are likely due to differences in patchiness. Annual species dominate according to the frequency method because they tend to have a more scattered distribution and show up in more quadrats. The creosote

bushes, on the other hand, are more widely scattered, but have dense cover in the places in which they occur. Lichens were absent from the macroplot. Exposed sand represented the largest cover class (Table 2), supporting our general impression that the soils at Yuma support a very scattered vascular plant community.

On 19 June 1995, we established 40 additional trampled plots. We were concerned that foot trampling was not severe enough to obtain an effect comparable to disturbance by military vehicles. Adjacent to the macroplot site (between it and the dirt access road) a four wheel drive vehicle was driven in circles and in criss-cross fashion over the ground to thoroughly disrupt the soil. These plots are numbered 26-65. They were not inoculated in the first inoculation series, but were inoculated the second time inoculations were performed. Inoculation was applied alternately, all odd-numbered plots in the second site receiving inoculum in February 1996.

All algal samples taken from plots were composite samples of ten 1 cm^3 surface samples taken randomly from the plots using a grid system with 400 defined points. We have found that composite samples are necessary to compensate for the spatial heterogeneity common in desert soils (Grondin and Johansen 1993).

Chlorophyll *a* concentrations were determined using the DMSO extraction method of Ronun and Galun (1984) as modified by Kasper (1994). Chlorophyll *a* analysis of soils from plots sampled for algal enumeration revealed very low algal biomass (Table 3). The mean for the site was 0.112 µg chl *a*/g soil, with values for plots ranging from undetectable to 0.340 µg chl *a*/g soil. These values are considerably below the means seen in studies at Dugway Proving Grounds, which were 8.02, 7.24, and 6.34 µg chl *a*/g soil for undisturbed, trampled, and burned plots, respectively. Statistical analysis of these and other data in the study were conducted using Statgraphics statistical software (STSC 1991). Multifactor analysis of variance on pretreatment values of chlorophyll *a* showed significant differences for blocks, but not for sites to be disturbed, or sites to be inoculated, giving some assurance that pre-existing differences between treatments were not present. We suspect that all these values are near detection limits, and thus the variability in the data may be due to precision errors rather than representative of real differences between plots. For this reason, we consider chlorophyll *a* data questionable, and have sought other methods to use in conjunction with chlorophyll *a* that are potentially more sensitive to differences in cyanobacterial and algal density, biomass, and recovery at Yuma Proving Grounds.

Plate counts, also called for in our original objectives, were not very reliable either. For the lowest dilution plate scorable (10^{-2} dilution), we often found 0 to 8 colony forming units (CFU's). Dilution plates are generally not considered accurate with fewer than 25 CFU's/plate. As unreliable as they may be, the CFU's/g dry soil are given for comparison with numbers obtained using the most probable number method discussed below (Table 3). There was no correlation between chlorophyll *a* data and plate count density data, which can be taken as further evidence that the methods are inappropriate for the Yuma soils.

Given the scarcity of algae at the Yuma Proving Grounds site, we did not perform epifluorescence counts. This is an exceedingly time intensive method that is only feasible when fairly high numbers of algae are in the soil. This is so because it is a direct count method, and thus provides no amplification through culturing as is obtained in the plate count and most probable number (MPN) methods. The MPN method was used to quantify the algae following disturbance and inoculation treatments in lieu of the epifluorescence method.

Algae were characterized through isolates from dilution plates, as well as some direct identification of cyanobacteria from moistened soils. The dominant cyanobacterial species were *Microcoleus vaginatus* and *Schizothrix calcicola*. A number of unusual chlorophytes were present in the soils of Yuma Proving Grounds. Seven of these are likely new to science (Table 4). The flora was distinctly different from that observed at Dugway Proving Grounds. Although the two sites share a number of cosmopolitan species, such as *Microcoleus vaginatus, Schizothrix calcicola, Nostoc punctiforme, Oscillatoria tenuis, Desmococcus vulgaris, Chlorella vulgaris, Myrmecia astigmatica, Hantzschia amphioxys,* and *Navicula mutica,* the new chlorophyte taxa, abundance of *Chlorococcum* and *Neochloris* species, absence of xanthophytes, and presence of unusual genera, such as *Chlorokybus* and *Characium,* separate the Yuma soils from those in the Great Basin Desert that we have studied. The Dugway soils had far more taxa as well, 79 taxa as compared to 37 taxa from Yuma Proving Grounds.

Soil characteristics for the macroplot and the second site exposed to vehicular traffic were determined for samples collected in February 1996. None of the samples were taken within the study plots. The two sites are very similar (Table 5). The vehicular disturbance site was notably lower in organic matter and nitrate. We suspect this is due to deeper mixing of the surface soil with subsurface soil by the four-wheel drive vehicle. Thus, the vehicular disturbance site had more dilution of the surface soil, which is relatively higher in organic matter and nitrate concentrations.

INOCULATION METHODOLOGY

We followed the protocols we developed for encapsulation of cyanobacteria for earlier inoculation studies at Dugway Proving Grounds. Since *Microcoleus vaginatus* was the most common cyanobacterium at Yuma Proving Grounds, and because we have had some success with growing this species in the past, we chose to inoculate the YPG plots with this species. *Microcoleus* was grown in Z-8 medium, a medium proven effective for growing soil cyanobacteria (Carmichael 1986). Cyanobacteria grow poorly in high light situations, so it was important to start the cultures off in small flasks placed in dimly lit incubators. Cultures were brought to higher volumes as density increased. By maintaining dense cultures, self shading could occur in the flasks, and this provided the necessary low light conditions. When a total of 12 liters of *Microcoleus* was in culture (4 3-liter spinner flasks), it was transferred to 250 liter capacity fiberglass tanks (18" diameter, 5' tall). These tanks were aerated using a pollution control air pump, which provided considerable lift and mixing in addition to the aeration.

Microcoleus vaginatus were encapsulated in 1.0% alginate (Kelgin F). This required 10 g alginate/liter liquid. Alginate is difficult to get into suspension. The alginate forms insoluble aggregates unless high shear mixing is used. We found that a kitchen blender worked well for both getting the alginate into suspension and breaking up large aggregates of the cyanobacterium. We processed 20-30 liters of cyanobacterial culture per batch. Once in solution, which had the consistency of soft gelatin, the alginate/cyanobacteria mix was poured into a drip tank with over one hundred drip points. The alginate mix dripped rapidly into a 2% calcium chloride solution, which caused the alginate to set into firm, small pellets.

After setting up (alginate needs at least 1 hour in the solution), the alginate pellets were transferred to unsterile media. This allowed the excess calcium chloride to diffuse out of the beads. If not removed, it was postulated that the calcium chloride could be deleterious to the cyanobacteria and would contribute to the conductivity of the soil, both undesirable outcomes. Unsterile Z-8 media was used to avoid osmotic shock and to provide some nutrients for the cyanobacteria, thus enhancing the chances of vigorous growth when the cyanobacteria were inoculated on the soil. Generally, the pellets were allowed to sit in unsterile media over night, although 4 hours was sufficient time for the calcium chloride to diffuse out of the pellets.

After sitting in the unsterile Z-8, pellets were removed from the media by pouring the liquid into a tank with a screened drain. The pellets were then placed on trays to dry, which usually took 1-3 days. After drying the pellets were kept refrigerated. Pellets were ground immediately before application to increase cyanobacterial escapability (Buttars et al. in review).

We wished the cyanobacterial amendment to have a uniform amount of cyanobacteria. This was done by weighing the amount of cyanobacteria that went into the solution. We decided on using 2.5 g algae/liter. Cultures were typically harvested when they had about 5 g algae/liter, so they usually had to be diluted before the alginate was added.

In February, 1995, all plots in the original macroplot designated as inoculation plots (Fig. 2) were inoculated with 100 g ground *Microcoleus* pellets/m^2. A year later, the plots exposed to vehicular disturbance were inoculated. In this set of plots (numbered 26-65) we inoculated all even-numbered plots.

In statistical analyses of our data we used two-tailed alternatives for the disturbance treatment, since we have seen some apparent stimulation of the *Microcoleus* populations in trampled soils. Blocks, a random variable of little interest, were also evaluated using a two-tailed alternative. For comparisons of inoculated and uninoculated plots, we used a one-tailed alternative, since we expected cyanobacterial biomass to either increase or stay the same in inoculated plots. Analysis of Variance (ANOVA) was used for all data sets. When data were clearly non-normal, we also used a nonparametric alternative, the Mann-Whitney test. For all tests, we selected $\alpha = 0.05$.

RESULTS OF INOCULATION STUDIES

The macroplot site was visited again on 23-24 May 1995, at which time all enumeration plots were sampled. We decided to use two algal enumeration methods. Chlorophyll *a* was determined because we have consistently used this method at other sites. We used the most probable number method because it is most accurate for low algal density soils. In an effort to increase our chances of getting significant results, we took samples from all 100 plots (both enumeration and sedimentation plots) for the MPN method.

The chlorophyll *a* data were promising. Trampled soils had significantly lower chlorophyll *a* contents than undisturbed soils. The blocks were also significant ($p=0.022$), indicating differences due to spatial heterogeneity were important. No interaction terms in the full model were significant. Inoculation was not a significant effect in the full model, although mean chlorophyll *a* was higher in both undisturbed and trampled plots that had been inoculated than the corresponding means for the uninoculated plots (Table 6). Because of the high number of zero readings, the chlorophyll *a* data were skewed. With non-normal data, often nonparametric methods are more powerful, and so we decided to use a Mann-Whitney test to examine comparisons between inoculation treatments within disturbance treatments. In the undisturbed plots, there was no significant difference between inoculated and uninoculated plots. However, in disturbed plots, the difference was significant, with inoculated plots having significantly ($p=0.040$) higher chlorophyll *a* values.

The most probable number data for the Yuma Proving Ground sites were similar to the chlorophyll *a* data. Promising trends were evidenced, but the inoculation effect was not significant. Two samples were lost during culturing, but 98 were processed and available for analysis. The effects of disturbance treatment (trampled, undisturbed), inoculation (inoculated, not inoculated), and blocking (25 blocks of 4 adjacent plots) were tested. According to multivariate ANOVA the undisturbed plots had significantly ($p=0.0008$) higher algal numbers than the trampled plots (Table 7). The inoculated soils had higher mean density than uninoculated soils, but the difference was not significant (Table 1). The blocks were also significant ($p=0.0021$), indicating differences due to spatial heterogeneity were important. No interaction terms in the full model were significant.

When a frequency histogram of all most probable number data was constructed, it was clear that the sample distribution was non-normal, being strongly skewed to the left (Fig. 3). A $\log_{10}$ transformation helped take some of the skewness out of the data (Fig. 4). When multivariate ANOVA was run on these log-transformed data the significance levels were higher for both disturbance and block treatments ($p<0.00005$), but were still not significant for the inoculation treatment. A nonparametric test, the Mann-Whitney test for two unpaired samples, was used as well, but without the blocking for the 25 blocks, the significance level for the disturbance treatment was $p=0.0077$, a value less significant than the p-value obtained when using untransformed data in multivariate ANOVA.

When a Mann-Whitney test was used to test the effects of inoculation on trampled plots, no significance was detected. A histogram (Fig. 5) of the

data used in this analysis shows the strong similarity in these distributions. It is interesting to note that the uninoculated plots had a higher incidence of zero abundances than inoculated plots (Fig. 5).

DISCUSSION

These data show the same trends as were found during similar studies at Dugway Proving Grounds. The trampling disturbance had significant deleterious effects on soil algal density and biomass. The difference was significant with chlorophyll *a* data, even though we were near detection levels using this methodology. The significance of the inoculation treatment was not apparent using a full ANOVA model. However, Mann-Whitney tests of data from just the disturbed plots indicated significantly higher chlorophyll *a* concentrations in inoculated plots.

The most probable number method is more sensitive than the chlorophyll *a* assay, and likewise showed significant differences between trampled and undisturbed plots, with undisturbed plots having higher algal densities. This method did not show a significant difference between inoculation treatments, although the mean density was slightly higher in inoculated plots.

Although it appears that we have had some success with the inoculation at Yuma Proving Ground, we are unsure that the significant differences observed are actually important. We plan to test the sites with wind to see if differences in erodibility exist within inoculated and uninoculated soils. The densities of algae in undisturbed plots in the sandy soil tested at Yuma Proving Ground are much lower than densities we have observed in any other site, and we are not sure that the algae in these soils play as significant a role as they do in soils of cooler and/or wetter deserts that have been studied by ourselves and other workers. Continued study of the site to determine the influence of soil cyanobacteria on erodibility and soil fertility are planned.

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Table 1. Sites at Yuma Proving Ground sampled for determination of algal species. Samples from these sites were collected May 13, 1994. Plate count data for YPG1-YPG6 are based on counts on BBM and Z-8.

Site	Location	Description
YPG1	32°51.622'N 114°24.69'W	Desert pavement, soil crusts between rocks. Near an anthill. The ants may have cleared out the rocks. Plate count data: $<3X10^3$ coccoids/g soil, $2.7X10^4$ cyanobacteria/g soil. About 75% of the cyanobacteria were *Schizothrix calcicola*. *Microcoleus vaginatus, Nostoc punctiforme, Schizothrix arenaria,* and *Hantzschia amphioxys* were also seen.
YPG2	32°50.992'N 114°24.322'W	Mars site, very desolate. Chemical crust. Plate count data: $<3X10^3$ algae/g soil. No algae were seen.
YPG3	32°48.927'N 114°23.434'W	Camp Laguna, noncrusted soil. Old camp for Patton's troops, abandoned 50 years ago. Plate count data: $2.0X10^4$ coccoids/g soil, $3.3X10^4$ cyanobacteria/g soil, $1.0X10^4$ diatoms/g soil. *Microcoleus vaginatus* predominated. *Schizothrix calcicola* was also present.
YPG4	32°48.927'N 114°23.434'W	Camp Laguna, crusted soil 100 feet away from site YPG3. Plate count data: $<3X10^3$ coccoids/g soil, $1.4X10^4$ cyanobacteria/g soil. *Microcoleus vaginatus* and *Schizothrix calcicola* were most common.
YPG5	32°50.373'N 114°22.111'W	Linear dunes near highway. Very coarse sandy soil. Plate count data: $<3X10^3$ algae/g soil. No algae seen.
YPG6	32°50.428'N 114°23.553'W	Legacy site, a site which has been highly disturbed by grading. Plate count data: $3X10^3$ coccoids/g soil, likely *Chlorella vulgaris*.
MACRO	32°51.661'N 114°22.281'W	Macroplot, sandy soil a little less coarse than YPG6. Near telephone pole number AC 99236. $4.1X10^2$ algae/g soil. *Microcoleus vaginatus* and *Schizothrix calcicola* were most common.

Table 2. Vascular plant characterization for Yuma Proving Ground. Frequency of occurrence (FREQ) was based on frequency in all 400 quadrats read in 100 plots. Percent relative frequency (REL FREQ) and percent cover (COVER) are also reported.

SPECIES	FREQ	REL FREQ	COVER
Abronia fragrans	12.00	2.39	.50
Achyronychia cooperi	4.25	.85	
Ambrosia dumosa	19.25	3.84	2.50
Brassica tournefortii	32.50	6.48	.25
Camissonia claviformis ssp. *yumae*	36.75	7.33	
Chorizanthe brevicornu	.25	.05	
Chorizanthe rigida	2.75	.55	
Chaenactis stevioides	57.00	11.37	1.50
Cryptantha micrantha	1.00	.20	
Cryptantha species	73.75	14.71	1.25
Dithyrea californica	15.00	2.99	
Draba verna	.50	.10	
Eremalche rotundifolia	.25	.05	
Eriogonum species	2.00	.40	
Eucrypta micrantha	.25	.05	
Hesperocallis undulata	3.50	.70	
Hilaria rigida	7.50	1.50	2.25
Larrea tridentata	8.50	1.69	4.75
Lepidium densiflorum	.75	.15	
Loeseliastrum schottii	19.25	3.84	
Lupinus concinnus	3.50	1.50	.25
Mentzelia albicaulis	34.25	6.83	
Nama demissum	8.75	1.74	
Oenothera species	8.75	1.74	
Palafoxia arida	9.00	1.79	
Pectocarya platycarpa	.75	.15	
Pectocarya setosa	1.50	.30	
Plantago ovata	44.00	8.77	.25
Rafinesquia californica	7.75	1.55	.25
Schismus barbatus	86.00	17.15	8.75
Tiquilia species	.25	.05	
VASCULAR PLANTS			22.50
LITTER			14.00
ROCK			.50
SAND			63.00

Table 3. Chlorophyll *a* (µg chl *a*/g dry soil) and algal density (CFU's/g soil) as estimated using the dilution plate technique for the 40 plots sampled in May 1994. Values represent samples taken prior to disturbance and inoculation, and are based on 3 replicate subsamples.

Plot	Disturbance	Treatment	Chl *a*	Block means	Density	Block means
4A	Undisturbed	Uninoculated	0.170	0.141	<100	<100
4B	Undisturbed	Inoculated	0.161		<100	
4C	Trampled	Inoculated	0.143		<100	
4D	Trampled	Uninoculated	0.089		<100	
5A	Undisturbed	Uninoculated	0.072	0.109	560	700
5B	Undisturbed	Inoculated	0.232		440	
5C	Trampled	Inoculated	0.072		1,560	
5D	Trampled	Uninoculated	0.063		220	
6A	Trampled	Uninoculated	0.340	0.199	<100	930
6B	Trampled	Inoculated	0.197		1,110	
6C	Undisturbed	Inoculated	0.152		1,890	
6D	Undisturbed	Uninoculated	0.107		670	
12A	Undisturbed	Uninoculated	0.241	0.185	1,110	760
12B	Undisturbed	Inoculated	0.170		1,670	
12C	Trampled	Inoculated	0.143		<100	
12D	Trampled	Uninoculated	0.188		220	
14A	Undisturbed	Inoculated	0.036	0.016	560	320
14B	Undisturbed	Uninoculated	0.009		330	
14C	Trampled	Uninoculated	0.018		<100	
14D	Trampled	Inoculated	0.000		330	
17A	Trampled	Uninoculated	0.206	0.089	<100	490
17B	Trampled	Inoculated	0.063		110	
17C	Undisturbed	Uninoculated	0.054		1,000	
17D	Undisturbed	Inoculated	0.036		780	
18A	Trampled	Inoculated	0.125	0.098	330	160
18B	Trampled	Uninoculated	0.063		220	
18C	Undisturbed	Inoculated	0.134		<100	
18D	Undisturbed	Uninoculated	0.072		<100	
20A	Trampled	Inoculated	0.063	0.067	110	<100
20B	Trampled	Uninoculated	0.143		<100	
20C	Undisturbed	Inoculated	0.027		110	
20D	Undisturbed	Uninoculated	0.036		110	
24A	Undisturbed	Uninoculated	0.089	0.123	<100	330
24B	Undisturbed	Inoculated	0.107		<100	
24C	Trampled	Inoculated	0.134		1,110	
24D	Trampled	Uninoculated	0.161		110	
25A	Undisturbed	Uninoculated	0.188	0.112	220	290
25B	Undisturbed	Inoculated	0.179		<100	
25C	Trampled	Inoculated	0.080		440	
25D	Trampled	Uninoculated	0.000		440	

Table 4. Species of soil algae collected from Yuma Proving Grounds (YPG), Yuma, Arizona. Taxa considered to be new to science are marked with an *. The full list of taxa at Dugway Proving Ground (DPG) is not given, but species present at both YPG and DPG are checked in the last column.

TAXA AT YPG	YPG1	YPG3	YPG4	MACRO	DPG
CYANOPHYTA					
Anabaena thermalis		X			
Microcoleus vaginatus	X	X	X	X	X
Nostoc punctiforme	X		X		X
Oscillatoria tenuis				X	X
Oscillatoria species				X	
Plectonema tomasinianum			X		X
Schizothrix arenaria	X				
Schizothrix calcicola	X	X	X	X	X
Scytonema ocellatum	X		X		X
Synechococcus cedrorum				X	
Synechocystis aquatilis				X	
Synechocytis pevalekii			X		
Tolypothrix species				X	X
CHLOROPHYTA					
Bracteacoccus minor var. *dispersa**	X				
Bracteacoccus pseudominor	X				
Bracteacoccus pseudominor var. *desertorum**	X				
Characium species*		X	X		
Chlorella emersonii				X	
Chlorella vulgaris		X	X		X
Chlorococcum arenosum			X		
Chlorococcum diplobionticum		X			
*Chlorococcum megagelatinosum**		X			
Chlorokybus atmosphyticus		X			
Chlorosarcinopsis aggregata	X	X			X
Chlorosarcinopsis aggregata var. *viridis**		X			
Chlorosarcinopsis auxotrophica	X				X
Chlorosarcinopsis gelatinosa	X	X	X		X
Chlorosarcinopsis minuta	X				
Chlorosarcinopsis species*	X	X			
Coccomyxa confluens		X			X
Neochloris pyrenoidosa		X			
Neochloris wimerii		X			
Pseudotetracystis species*		X			
Spongiochloris elanoesis		X			
BACILLARIOPHYTA					
Hantzschia amphioxys	X	X			X
Navicula mutica		X			X
Navicula mutica var. *cohnii*		X			X

Table 5. Soil characteristics for the two test sites at Yuma Proving Ground, the macroplot (MACRO) and the vehicular disturbance site (VEHIC). Means and standard errors are reported for each listed parameter.

Parameter	MACRO	VEHIC
Percent sand	89.66±.14	88.48±1.04
Percent silt	4.12±.19	5.33±.38
Percent clay	6.20±.10	6.20±.72
Percent organic matter	.19±.05	.13±.01
pH	8.16±.04	8.14±.05
Nitrate-N (ppm)	3.20±.56	2.21±.06
Phosphorus (ppm)	1.84±.12	2.63±.35
Potassium (ppm)	84.48±5.02	86.40±10.52
Calcium (ppm)	82.37±5.98	73.70±3.12
Magnesium (ppm)	8.99±.60	8.61±.94
Sodium (ppm)	9.98±1.06	9.76±.68
SAR	.28±.02	.29±.02
EC x 10^3	.52±.03	.49±.01

Table 6. Results of chlorophyll *a* analysis for 40 samples collected post disturbance and inoculation, 23-24 May 1995. Standard errors are based on sum of squares from multifactor ANOVA (using both main effects and interaction sources of variation). Disturbed plots had significantly higher chlorophyll *a* concentrations than undisturbed plots. Within the disturbed plots only, a Mann-Whitney test indicated significantly higher chlorophyll *a* concentrations in the inoculated plots.

	Inoculated	Uninoculated	Mean (disturbance)
Undisturbed	0.173±0.075	0.071±0.075	0.122±0.053
Trampled	0.077±0.075	0.009±0.075	0.043±0.053
Mean (Inoculation)	0.125±0.053	0.040±0.053	0.083±0.038

Table 7. Results of most probable number analysis for 100 samples collected post disturbance and inoculation, 23-24 May 1995. Mean numbers of CFU's/g soil are reported with standard error. Standard errors are based on sum of squares from multifactor ANOVA (using both main effects and interaction sources of variation). Undisturbed plots had significantly higher densities of algae than trampled plots. Blocks were a significant effect, but means are not shown since this is a random factor. All interaction terms were not significant.

	Inoculated	Uninoculated	Mean (disturbance)
Undisturbed	553±102	494±102	524±72
Trampled	221±102	229±102	212±74
Mean (Inoculation)	387±73	361±73	374±52

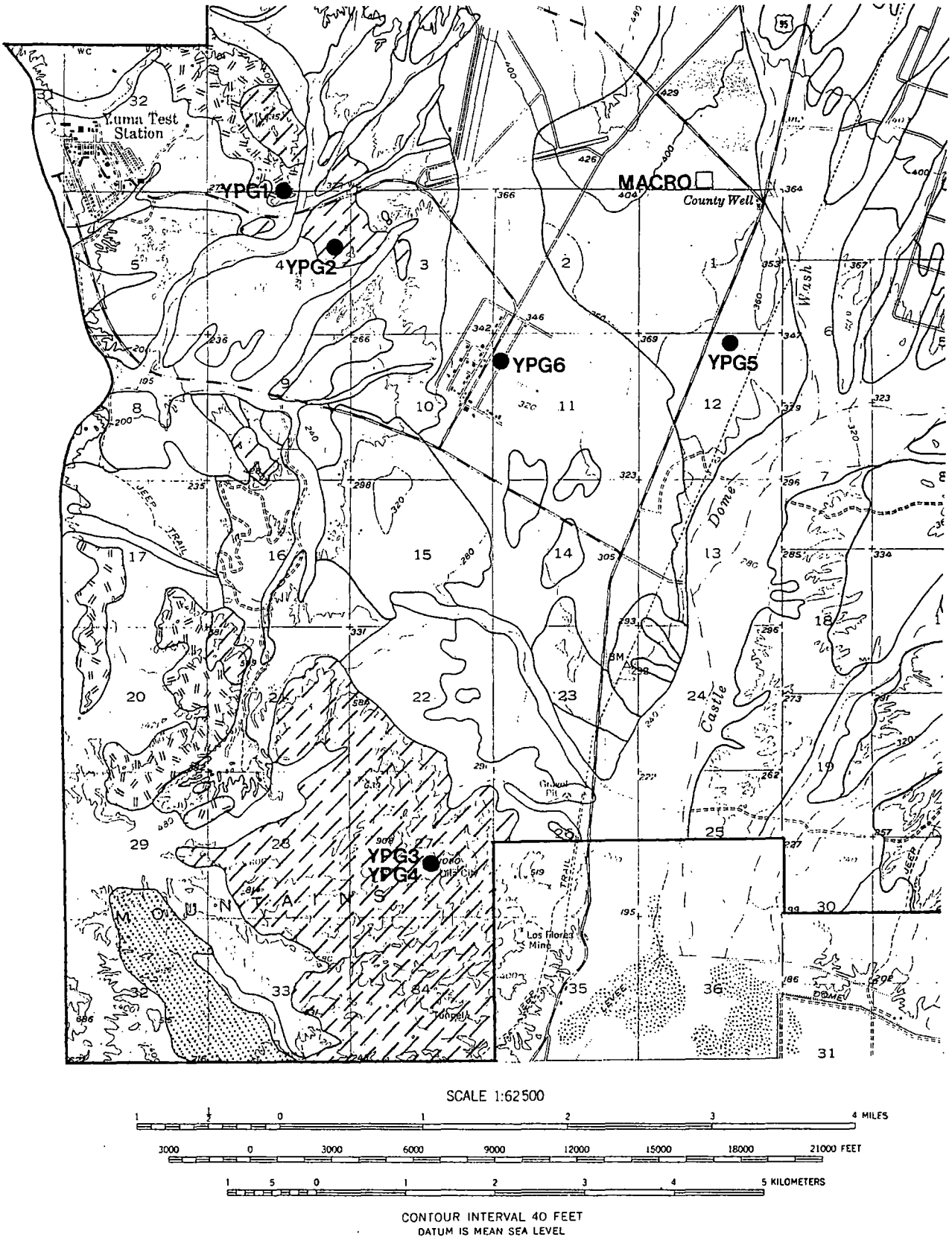

Figure 1. Map of study plots at Yuma Proving Grounds. See Table 1 for descriptions of sites.

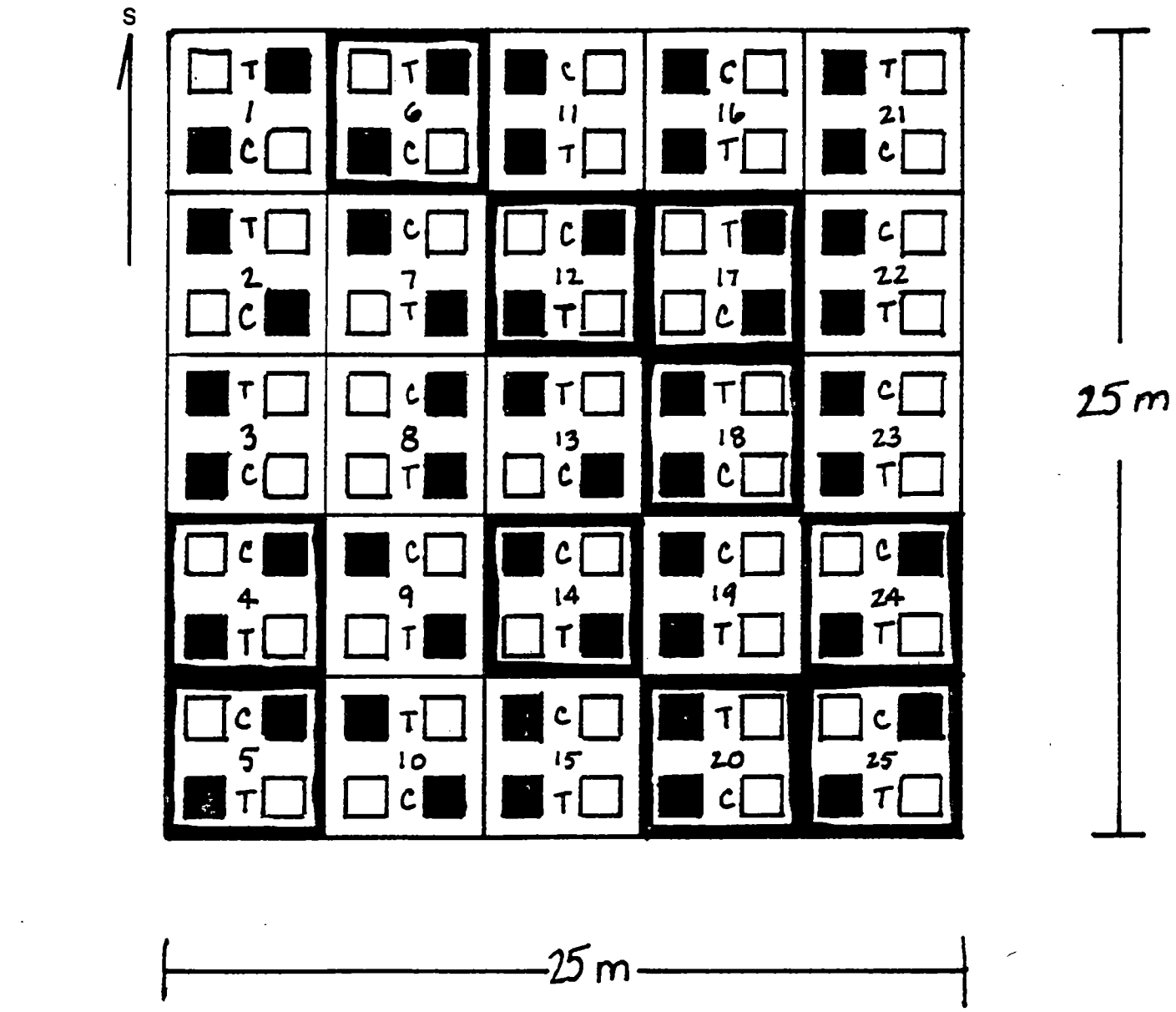

Figure 2. Map of Macroplot. The top of the plot is the edge nearest to the access road. Sampled blocks have thicker margins.

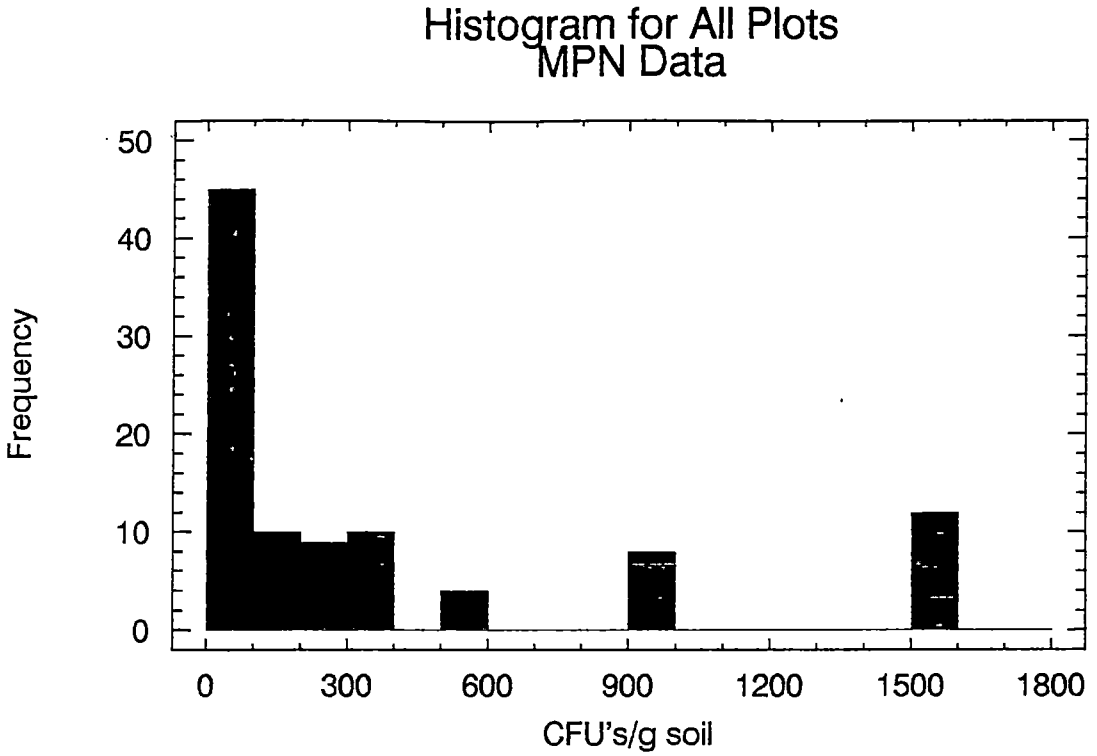

Figure 3. Histogram of most probable number data for all plots sampled in May 1995. The data are highly skewed and distinctly non-normal. Numbers reported are in colony forming units (CFU's) per gram dry soil.

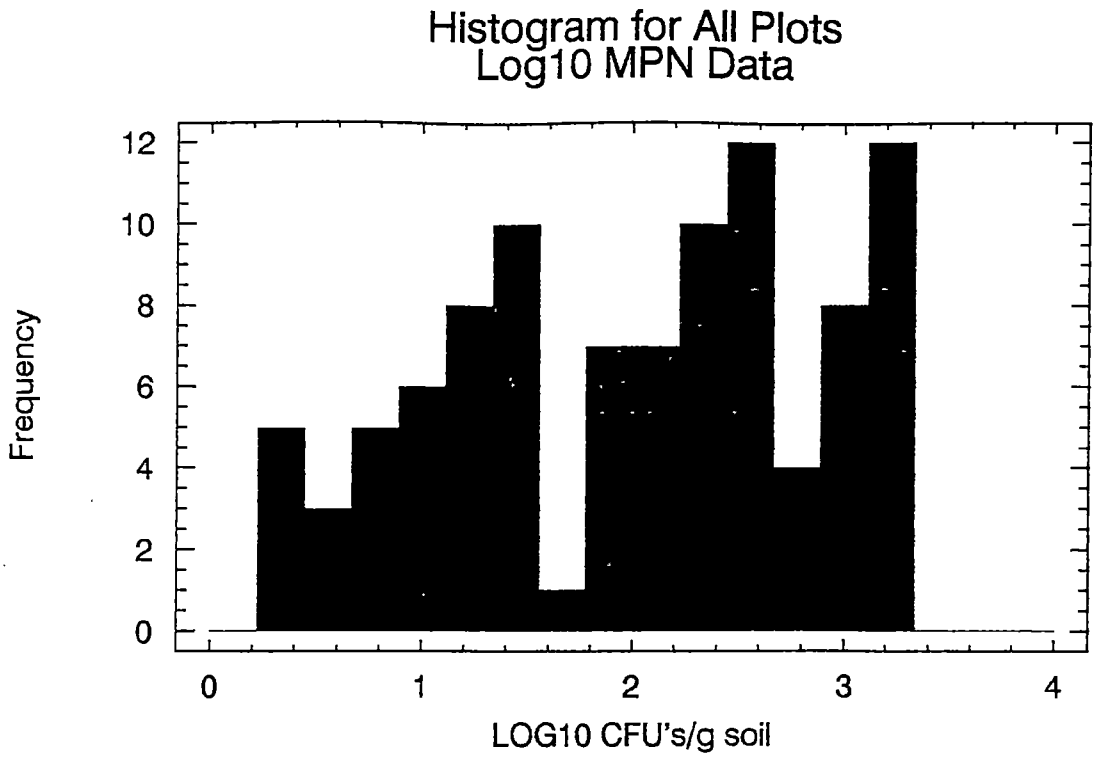

Figure 4. Histogram of $\log_{10}$ transformed most probable number data for all plots sampled in May 1995. The data are less skewed but still non-normal.

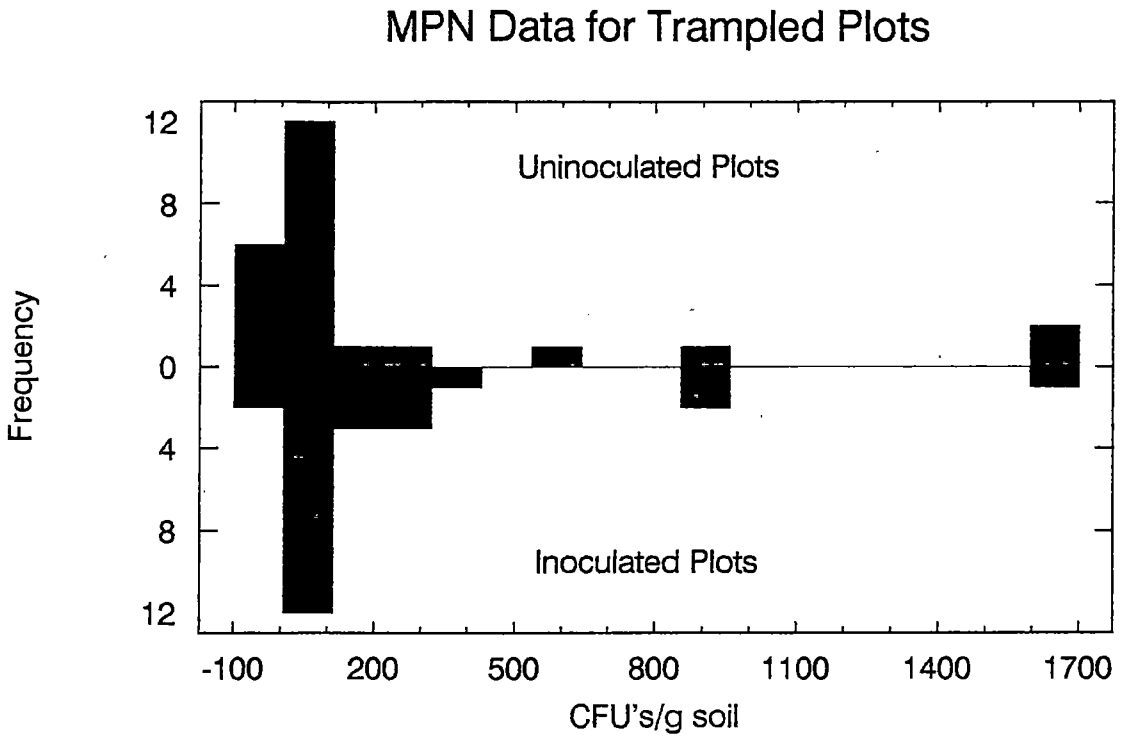

Figure 5.. Histogram of most probable number data for trampled plots sampled in May 1995, showing separate distributions for inoculated and uninoculated plots. Again, the data are highly skewed and distinctly non-normal. Numbers reported are in colony forming units (CFU's) per gram dry soil. Note the higher incidence of zero abundances in the uninoculated plots.